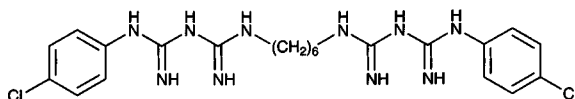


Chlorhexidine



Molecular formula: $C_{22}H_{30}Cl_2N_{10}$

Molecular weight: 505.45

CAS Registry No.: 55-56-1, 3697-42-5 (dihydrochloride),
18472-51-0 (gluconate), 77146-42-0 (phosphanilate)

Merck Index: 2140

SAMPLE

Matrix: blood, urine

Sample preparation: 500 μ L Serum or urine + 50 μ L 50 μ g/mL chlorpheniramine in water + 100 μ L 2 M NaOH + 2.5 mL chloroform:isopropanol 95:5, shake vigorously for 20 min, centrifuge at 700 g for 5 min. Remove the organic layer and add it to 100 μ L 50 mM sulfuric acid, mix vigorously on a shaker for 10 min, inject a 20 μ L aliquot of the aqueous layer.

HPLC VARIABLES

Column: 300 \times 3.9 10 μ m μ Bondapak C18

Mobile phase: MeOH:water 70:30 containing 5 mM sodium heptanesulfonate

Flow rate: 1.2

Injection volume: 20

Detector: UV 260

CHROMATOGRAM

Retention time: 8.0

Internal standard: chlorpheniramine (4.9)

Limit of detection: 20 ng/mL

OTHER SUBSTANCES

Simultaneous: p-chloroaniline

Noninterfering: benzyl alcohol, benzoic acid, phosphanilic acid

KEY WORDS

serum

REFERENCE

Brougham, L.R.; Cheng, H.; Pittman, K.A. Sensitive high-performance liquid chromatographic method for the determination of chlorhexidine in human serum and urine, *J. Chromatogr.*, **1986**, 383, 365–373.

SAMPLE

Matrix: blood, urine

Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μ L MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μ L aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200–350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18

Column: 250 \times 4.6 5 μ m Symmetry C8 (Waters)

Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A: B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.

Column temperature: 30

Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)

Injection volume: 10-30

Detector: UV 200.5

CHROMATOGRAM

Retention time: 13.532

KEY WORDS

whole blood

REFERENCE

Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, 763, 149–163.

SAMPLE

Matrix: bulk

Sample preparation: Prepare a 10 mg/mL solution in MeCN:100 mM pH 5.0 ammonium acetate 20:80.

HPLC VARIABLES

Column: 150 × 4.6 3 µm Nucleosil C18 (A) or 250 × 4.6 5 µm Zorbax RX-C8 (B)

Mobile phase: A was MeCN. B was 100 mM pH 5.0 ammonium acetate. Gradient. A:B from 20:80 to 25:75 over 25 min, to 40:60 over 25 min, to 50:50 over 40 min, return to initial conditions, re-equilibrate for 10 min.

Column temperature: 30

Flow rate: 1

Injection volume: 20

Detector: UV 230

CHROMATOGRAM

Retention time: 45-50 (A) or 45 (B)

OTHER SUBSTANCES

Simultaneous: degradation products

KEY WORDS

comparison of columns

REFERENCE

Doub,W.H.; Ruhl,D.D.; Hart,B.; Mehelic,P.R.; Revelle,L.K. Gradient liquid chromatographic method for the determination of chlorhexidine and its degradation products in bulk material, *J.AOAC Int.*, **1996**, 79, 636–639.

SAMPLE

Matrix: formulations

Sample preparation: Weigh out 1.25 g ground pastilles, add 50 mL mobile phase, stir mechanically until dissolved, inject a 50 µL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 µm µBondapak C18

Mobile phase: MeCN:100 mM pH 5 potassium phosphate buffer containing 5.9 g/L NaCl and 30 mM tetrabutylammonium hydrogen sulfate

Flow rate: 2

Injection volume: 50

Detector: UV 294

CHROMATOGRAM

Retention time: 10

OTHER SUBSTANCES

Simultaneous: degradation products, n-butyl p-aminobenzoic acid, p-chloroaniline, tetracaine

KEY WORDS

pastilles

REFERENCE

Bauer,M.; Degude,C.; Mailhe,L. Simultaneous determination of chlorhexidine, tetracaine and their degradation products by ion-pair liquid chromatography, *J.Chromatogr.*, **1984**, *315*, 457–464.

SAMPLE

Matrix: saliva

Sample preparation: Centrifuge saliva at 200 g, remove a 200 μ L aliquot, add 200 μ L 5 M KOH, vortex for 30 s, add 2 mL dichloromethane, shake vigorously on a horizontal shaker for 10 min, centrifuge at 200 g for 10 min. Remove the organic phase and evaporate it under a stream of nitrogen, reconstitute with 100 μ L mobile phase, inject a 60 μ L aliquot.

HPLC VARIABLES

Column: 150 \times 4.6 5 μ m Ultrasphere ODS C18

Mobile phase: MeCN:50 mM sodium acetate + 5 mM heptanesulfonic acid 40:60 adjusted to pH 5 with glacial acetic acid

Flow rate: 1

Injection volume: 60

Detector: UV 260

CHROMATOGRAM

Retention time: 6.1

Limit of detection: 50 ng/mL

OTHER SUBSTANCES

Noninterfering: erythromycin, lidocaine, tetracycline

REFERENCE

Lam,Y.W.F.; Chan,D.C.N.; Rodriguez,S.Y.; Lintakoon,J.H.; Lam,T.-H. Sensitive high-performance liquid chromatographic assay for the determination of chlorhexidine in saliva, *J.Chromatogr.*, **1993**, *612*, 166–171.

SAMPLE

Matrix: saliva

Sample preparation: Collect sample on Periopaper strip (filter paper), add paper to 100 μ L MeCN:water:glacial acetic acid 55:44.8:0.2 containing 7 mM sodium lauryl sulfate and 2 μ g/mL benzethonium, vortex for 1 min, sonicate for 20 min, vortex, inject a 50 μ L aliquot.

HPLC VARIABLES

Column: 100 \times 2.1 5 μ m C18 ODS-B Exsil (HiChrome)

Mobile phase: MeCN:0.2% acetic acid 55:45 containing 5 mM sodium lauryl sulfate
Flow rate: 0.5
Injection volume: 50
Detector: UV 254

CHROMATOGRAM

Retention time: 5
Internal standard: benzethonium (7)
Limit of detection: 2000 ng/mL

KEY WORDS

narrow-bore

REFERENCE

Medlicott,N.J.; Ferry,D.G.; Tucker,I.G.; Rathbone,M.J.; Holborow,D.W.; Jones,D.S. High performance liquid chromatographic (HPLC) assay for the determination of chlorhexidine in saliva film, *J.Liq.Chromatogr.*, **1994**, *17*, 1605–1620.

SAMPLE

Matrix: saliva

Sample preparation: 200 μ L Saliva + 400 μ L 4.5 M NaOH + 400 μ L MeCN, vortex for 1 min, centrifuge for 1 min at 14000 g. Remove 200 μ L of the organic phase and add it to 370 μ L buffer, mix, inject a 20 μ L aliquot. (Buffer was 100 mM Na_2HPO_4 containing 5 mM 1-heptanesulfonic acid and 50 mM triethylamine, pH adjusted to 2.5 with phosphoric acid.)

HPLC VARIABLES

Column: 125 \times 4.5 μ m LiChrospher 100 RP-18
Mobile phase: MeCN:buffer 35:65 (Buffer was 100 mM Na_2HPO_4 containing 5 mM 1-heptanesulfonic acid and 50 mM triethylamine, pH adjusted to 2.5 with phosphoric acid.)
Injection volume: 20
Detector: UV 260

CHROMATOGRAM

Retention time: 2.10
Limit of quantitation: 500 ng/mL

KEY WORDS

pharmacokinetics

REFERENCE

Pesonen,T.; Holmalahti,J.; Pohjola,J. Determination of chlorhexidine in saliva using high-performance liquid chromatography, *J.Chromatogr.B*, **1995**, *665*, 222–225.

SAMPLE

Matrix: solutions

Sample preparation: 9.5 mL Contact lens solution + 0.5 mL 3 mg/mL methylparaben, inject a 20 μ L aliquot.

HPLC VARIABLES

Guard column: 7 μ m Nucleosil C18 pre-column
Column: 7 μ m Nucleosil C18
Mobile phase: MeOH:100 mM KH_2PO_4 adjusted to pH 3.5 with phosphoric acid 60:40
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM

Retention time: 14.9 (chlorhexidine gluconate)

Internal standard: methyl paraben (7.8)

Limit of detection: 200 ng

OTHER SUBSTANCES

Simultaneous: thimerosal, thiosalicylic acid

KEY WORDS

stability-indicating; contact lens solutions

REFERENCE

Hu,O.Y.-P.; Wang,S.-Y.; Fang,Y.-J.; Chen,Y.-H.; King,M.-L. Simultaneous determination of thimerosal and chlorhexidine in solutions for soft contact lenses and its applications in stability studies, *J.Chromatogr.*, **1990**, 523, 321–326.

SAMPLE

Matrix: urine

Sample preparation: Direct injection of a 20 μ L aliquot.

HPLC VARIABLES

Column: 250 \times 5 10 μ m Partisil silica

Mobile phase: MeOH:2 M ammonia:1 M ammonium nitrate 90:5:5

Flow rate: 3

Injection volume: 20

Detector: UV 260

CHROMATOGRAM

Retention time: 5.5

Limit of quantitation: 1000 ng/mL

KEY WORDS

normal phase; use a 50 mm long column of 27-44 μ m silica between pump and injection valve to saturate mobile phase with silica

REFERENCE

Wainwright,P.; Cooke,M. Direct determination of chlorhexidine in urine by high-performance liquid chromatography, *Analyst*, **1986**, 111, 1343–1344.

Chlormadinone

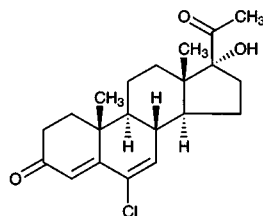
Molecular formula: $C_{21}H_{27}ClO_3$

Molecular weight: 362.90

CAS Registry No.: 1961-77-9, 302-22-7 (acetate)

Merck Index: 2152

Lednicer No.: 1 181



SAMPLE

Matrix: blood, urine

Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μ L MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μ L aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18

Column: 250 \times 4.6 5 μ m Symmetry C8 (Waters)

Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A: B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.

Column temperature: 30

Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)

Injection volume: 10-30

Detector: UV 283.1

CHROMATOGRAM

Retention time: 24.11

KEY WORDS

whole blood

REFERENCE

Gaillard, Y.; Pépin, G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J. Chromatogr. A*, **1997**, 763, 149-163.

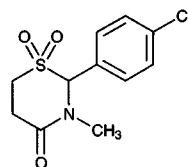
Chlormezanone

Molecular formula: C₁₁H₁₂ClNO₃S

Molecular weight: 273.74

CAS Registry No.: 80-77-3

Merck Index: 2155



SAMPLE

Matrix: blood

Sample preparation: 1 mL plasma + 100 mg ammonium sulfate + 2 mL ethyl acetate, vortex for 2 min, sonicate for 3 min, centrifuge at 6000 rpm for 10 min. Remove 1 mL of the supernatant and evaporate it to dryness under vacuum, reconstitute the residue in 500 μ L mobile phase, inject a 20-40 μ L aliquot.

HPLC VARIABLES

Guard column: 30 \times 4 7.5 μ m Nucleosil C18

Column: 120 \times 4 7.5 μ m Nucleosil C18

Mobile phase: MeOH:water:85% orthophosphoric acid 50:50:1

Flow rate: 1

Injection volume: 20-40

Detector: UV 228

CHROMATOGRAM

Retention time: 3.37

Limit of detection: 100 ng/mL

Limit of quantitation: 500 ng/mL

OTHER SUBSTANCES

Simultaneous: acetaminophen

Noninterfering: codeine

KEY WORDS

plasma; pharmacokinetics

REFERENCE

Ali,S.L.; Blume,H. Determination of chlormezanone in human plasma after administration of chlormezanone formulations, *Arzneimittelforschung*, **1987**, 37, 1396-1399.

SAMPLE

Matrix: blood

Sample preparation: 200 μ L Plasma + 300 μ L MeCN, mix, allow to stand for 10 min, mix, centrifuge for 5 min, inject an aliquot of the supernatant

HPLC VARIABLES

Guard column: 20 \times 4 5 μ m Nucleosil C18

Column: 120 \times 4 5 μ m Nucleosil C18

Mobile phase: MeCN:water:85% orthophosphoric acid 35:65:0.1

Flow rate: 1.5

Injection volume: 20

Detector: UV 225

CHROMATOGRAM

Retention time: 4.12

Limit of detection: 50 ng/mL

Limit of quantitation: 200 ng/mL

OTHER SUBSTANCES

Extracted: metabolites

KEY WORDS

plasma; pharmacokinetics

REFERENCE

Koppel,C.; Kristinsson,J.; Wagemann,A.; Tenczer,J.; Martens,F. Chlormezanone plasma and blood levels in patients after single and repeated oral doses and after suicidal drug overdose, *Eur.J.Drug Metab.Pharmacokinet.*, **1991**, 16, 43–47.

SAMPLE

Matrix: blood

Sample preparation: 2 mL Whole blood or plasma + 2 mL buffer + 5 mL chloroform: isopropanol:n-heptane 60:14:26, shake gently horizontally for 10 min, centrifuge at 2800 g for 10 min. Remove the lower organic layer and evaporate it to dryness under vacuum at 45°, reconstitute the residue in 100 µL mobile phase, centrifuge at 2800 g for 5 min, inject a 50 µL aliquot of the supernatant. (Buffer was saturated ammonium chloride solution 25% diluted with water, adjusted to pH 9.5 with 25% ammonia solution.)

HPLC VARIABLES

Column: 300 × 3.9 4 µm NovaPack C18

Mobile phase: MeOH:THF:buffer 65:5:30 (Buffer was 0.68 g/L (10 mM (sic)) KH₂PO₄ adjusted to pH 2.6 with concentrated orthophosphoric acid.) (At the end of each session wash the column with water for 1 h and MeOH for 1 h, re-equilibrate for 30 min.)

Column temperature: 30

Flow rate: 0.8

Injection volume: 50

Detector: UV 223

CHROMATOGRAM

Retention time: 3.40

Limit of detection: <120 ng/mL

KEY WORDS

whole blood; plasma; interferences may occur—compounds(all of which are extracted) elute in this order tenoxicam; iproniazid; methocarbamol; methotrexate; caffeine; nialamide; colchicine; cytarabine; benzoylecgonine; acetaminophen; diazoxide; dacarbazine; sulfinpyrazole; flumazenil; sulpride; morphine; atenolol; toloxatone; terbutaline; albuterol; phenobarbital; ranitidine; tiapride; phenol; chlormezanone; aspirin; metformin; ritodrine; codeine; sultopride; amisulpride; naltrexone; lisinopril; benzocaine; nizatidine; nalorphine; mephenesin; naloxone; sotalol; carteolol; procainamide; carbamazepine; bromazepam; nalbuphine; nadolol; procarbazine; dihydralazine; omeprazole; strychnine; acebutolol; glutethimide; chlorpropamide; glipizide; triazolam; prazosin; flunitrazepam; clonazepam; metoclopramide; melphalan; estazolam; tolbutamide; ephedrine; clonidine; pindolol; clobazam; minoxidil; disopyramide; nitrazepam; dextromethorphan; tofisopam; zopiclone; debrisoquine; sulindac; alprazolam; cycloguanil; lorazepam; methaqualone; ketamine; piroxicam; metoprolol; nifedipine; quinine; mephentermine; prilocaine; pentazocine; oxazepam; tiaprofenic acid; quinidine; celiprolol; ajmaline; yohimbine; lidocaine; secobarbital; viloxazine; mepivacaine; meperidine; doxylamine; labetalol; temazepam; amodiaquine; benperidol; droperidol; hydroxychloroquine; zolpidem; ketoprofen; alminoprofen; cicletanine; moclobemide; chloroquine; cocaine; timolol; nomifensine; ticlopidine; acenocoumarol; videsine; mexiletine; dipyridamole; trazodone; pipamperone; pyrimethamine; benazepril; vincristine; metapramine; chlordiazepoxide; oxprenolol; warfarin; clorazepate; flecainide; phenacyclidine; thiopental; fenfluramine; metipranolol; triprolidine; naproxen; buprenorphine; verapamil; buspirone; tianeptine; midazolam; bupivacaine; carbinoxamine; loperazolam; cetirizine; chlorpheniramine; moperone; cibenzoline; medifoxamine; astemizole; vinblastine; nicardipine; bisoprolol; diltiazem; glibornuride; reserpine;

aconitine; nitrendipine; diazepam; mianserin; ramipril; haloperidol; tetracaine; alprenolol; aceprometazine; glibenclamide; chlorophenacinone; doxepin; nimodipine; diphenhydramine; cyclizine; histapyrrodine; phenylbutazone; demexiptiline; clozapine; proguanil; trifluoperidol; medazepam; cyamemazine; bumadizone; suriclone; propranolol; acepromazine; dothiepin; dextromoramide; fenoprofen; dextropropoxyphene; loxapine; betaxolol; propafenone; promethazine; thiopropazine; methadone; amoxapine; quinupramine; opipramol; cyproheptadine; brompheniramine; mefenidramine; protriptyline; flurbiprofen; tetrazepam; zorubicin; prazepam; alimemazine; loperamide; imipramine; desipramine; levomepromazine; hydroxyzine; niflumic acid; penbutolol; fluvoxamine; pimozone; daunorubicin; indomethacin; maprotiline; tropatenine; etodolac; fluoxetine; amitriptyline; nortriptyline; tioclomarol; diclofenac; mefloquine; trimipramine; chlorambucil; lidoflazine; ibuprofen; floctafenine; alpidem; loratadine; chlorpromazine; clomipramine; carpipramine; thioridazine; fentiazac; clemastine; mefenamic acid; fluphenazine; prochlorperazine; penfluridol; bepridil; terfenadine; trifluoperazine

REFERENCE

Tracqui, A.; Kintz, P.; Mangin, P. Systematic toxicological analysis using HPLC/DAD, *J. Forensic Sci.*, **1995**, *40*, 254–262.

SAMPLE

Matrix: blood, urine

Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 µL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) µL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200–350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18

Column: 250 × 4.6 5 µm Symmetry C8 (Waters)

Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A: B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.

Column temperature: 30

Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)

Injection volume: 10–30

Detector: UV 200.5

CHROMATOGRAM

Retention time: 15.493

KEY WORDS

whole blood

REFERENCE

Gaillard, Y.; Pépin, G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J. Chromatogr. A*, **1997**, *763*, 149–163.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 5 µm Supelcosil LC-DP (A) or 250 × 4.5 µm LiChrospher 100 RP-8 (B)

Mobile phase: MeCN:0.025% phosphoric acid:buffer 25:10:5 (A) or 60:25:15 (B) (Buffer was 9 mL concentrated phosphoric acid and 10 mL triethylamine in 900 mL water, adjust pH to 3.4 with dilute phosphoric acid, make up to 1 L.)

Flow rate: 0.6

Injection volume: 25

Detector: UV 229

CHROMATOGRAM

Retention time: 6.03 (A), 5.33 (B)

OTHER SUBSTANCES

Also analyzed: acebutolol, acepromazine, acetaminophen, acetazolamide, acetophenazine, albuterol, alprazolam, amitriptyline, amobarbital, amoxapine, antipyrine, atenolol, atropine, azatadine, baclofen, benzocaine, bromocriptine, brompheniramine, brotizolam, bupivacaine, buspirone, butabarbital, butalbital, caffeine, carbamazepine, cetirizine, chlorcyclizine, chlordiazepoxide, chloroquine, chlorpheniramine, chlorpromazine, chlorpropamide, chlorprothixene, chlorthalidone, chlorzoxazone, cimetidine, cisapride, clomipramine, clonazepam, clonidine, clozapine, cocaine, codeine, colchicine, cyclizine, cyclobenzaprine, dantrolene, desipramine, diazepam, diclofenac, diflunisal, diltiazem, diphenhydramine, diphenidol, diphenoxylate, dipyrindamole, disopyramide, dobutamine, doxapram, doxepin, droperidol, encainide, ethidium bromide, ethopropazine, fenopropfen, fentanyl, flavoxate, fluoxetine, fluphenazine, flurazepam, flurbiprofen, fluvoxamine, furosemide, glutethimide, glyburide, guaifenesin, haloperidol, homatropine, hydralazine, hydrochlorothiazide, hydrocodone, hydromorphone, hydroxychloroquine, hydroxyzine, ibuprofen, imipramine, indomethacin, ketoconazole, ketoprofen, ketorolac, labetalol, levorphanol, lidocaine, loratadine, lorazepam, lovastatin, loxapine, mazindol, mefenamic acid, meperidine, mephentoin, mepivacaine, mesoridazine, metaproterenol, methadone, methdilazine, methocarbamol, methotrexate, methotrimeprazine, methoxamine, methyl-dopa, methylphenidate, metoclopramide, metolazone, metoprolol, metronidazole, midazolam, moclobemide, morphine, nadolol, nalbuphine, naloxone, naphazoline, naproxen, nifedipine, nizatidine, norepinephrine, nortriptyline, oxazepam, oxycodone, oxymetazoline, paroxetine, pemoline, pentazocine, pentobarbital, pentoxifylline, perphenazine, pheniramine, phenobarbital, phenol, phenolphthalein, phentolamine, phenylbutazone, phenyltoloxamine, phenytoin, pimizide, pindolol, piroxicam, pramoxine, prazepam, prazosin, probenecid, procainamide, procaine, prochlorperazine, procyclidine, promazine, promethazine, propafenone, propantheline, propiomazine, propofol, propranolol, protriptyline, quazepam, quinidine, quinine, racemethorphan, ranitidine, remoxipride, risperidone, salicylic acid, scopolamine, secobarbital, sertraline, sotalol, spironolactone, sulfapyrazone, sulindac, temazepam, terbutaline, terfenadine, tetracaine, theophylline, thiethylperazine, thiopental, thioridazine, thiothixene, timolol, tocinide, tolbutamide, tolmetin, trazodone, triamterene, triazolam, trifluoperazine, triflupromazine, trimeprazine, trimethoprim, trimipramine, verapamil, warfarin, xylometazoline, yohimbine, zopiclone

KEY WORDS

also details of plasma extraction

REFERENCE

Koves, E.M. Use of high-performance liquid chromatography-diode array detection in forensic toxicology, *J. Chromatogr. A*, **1995**, 692, 103–119.

Chloroprocaine

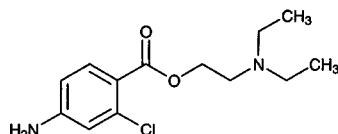
Molecular formula: C₁₃H₁₉ClN₂O₂

Molecular weight: 270.76

CAS Registry No.: 133-16-4, 3858-89-7 (HCl)

Merck Index: 2210

Lednicer No.: 1 11



SAMPLE

Matrix: bulk, formulations

Sample preparation: 100 mg Bulk drug or formulation containing 100-120 mg drug + 10 mL 1 (bulk) or 4 (formulations) mg/mL benzoic acid in MeOH:water 50:50 + 20 mL 5 mg/mL p-nitroacetophenone in MeOH, make up to 100 mL with water, inject an aliquot.

HPLC VARIABLES

Column: 300 × 4 10 μm μBondapak C18

Mobile phase: MeCN:MeOH:water:glacial acetic acid 20:5:74:1 containing 0.05-0.08% sodium 1-heptanesulfonate, pH 3.1

Flow rate: 2

Injection volume: 5

Detector: UV 278

CHROMATOGRAM

Retention time: 9

Internal standard: benzoic acid (6), p-nitroacetophenone (12)

OTHER SUBSTANCES

Simultaneous: 4-amino-2-chlorobenzoic acid, impurities

REFERENCE

Menon,G.; Norris,B.; Webster,J. Simultaneous determination of chloroprocaine hydrochloride and its degradation product 4-amino-2-chlorobenzoic acid in bulk drug and injection solutions by high-performance liquid chromatography, *J.Pharm.Sci.*, **1984**, 73, 251-253.

SAMPLE

Matrix: perfusate

Sample preparation: Adjust pH of 5-10 mL perfusate to 5 with 180 μL 2.5 M HCl, extract twice with an equal volume of ethyl acetate. Combine the organic layers, add 1 mL water, evaporate them to 1 mL under vacuum, inject a 20 μL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 μm μBondapak C18

Mobile phase: MeOH:water containing 30 μL/L triethylamine, adjusted to pH 2.3 with phosphoric acid 20:80

Flow rate: 1.5

Injection volume: 20

Detector: UV 290

CHROMATOGRAM

Retention time: 9.2

OTHER SUBSTANCES

Extracted: 2-chloro-4-aminobenzoic acid, 4-acetamidohippuric acid

KEY WORDS

rabbit; chinchilla; pharmacokinetics

REFERENCE

Henrikus,B.M.; Kampffmeyer,H.G. Ester hydrolysis and conjugation reactions in intact skin and skin homogenate, and by liver esterase of rabbits, *Xenobiotica*, **1992**, 22, 1357–1366.

SAMPLE

Matrix: solutions

Sample preparation: Dissolve in MeOH:water 1:1 at a concentration of 50 µg/mL, inject a 10 µL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 µm µBondapak C18

Mobile phase: MeOH:acetic acid:triethylamine:water 30:1.5:0.5:68

Flow rate: 1.5

Injection volume: 10

Detector: UV

CHROMATOGRAM

Retention time: k' 0.92

REFERENCE

Roos,R.W.; Lau-Cam,C.A. General reversed-phase high-performance liquid chromatographic method for the separation of drugs using triethylamine as a competing base, *J.Chromatogr.*, **1986**, 370, 403–418.

Chloropyramine

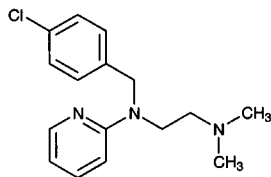
Molecular formula: C₁₆H₂₀ClN₃

Molecular weight: 289.81

CAS Registry No.: 59-32-5, 6170-42-9 (HCl)

Merck Index: 2214

Lednicer No.: 1 402



SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 150 × 4.6 12 μm 1-myristoyl-2-[(13-carboxyl)-tridecoyl]-sn-3-glycerophosphocholine chemically bonded to silica (Regis)

Mobile phase: MeCN:100 mM pH 7.0 phosphate buffer 20:80

Flow rate: 1

Detector: UV 254

CHROMATOGRAM

Retention time: k' 21.38

OTHER SUBSTANCES

Also analyzed: acebutolol, alprenolol, antazoline, atenolol, betaxolol, bisoprolol, bopindolol, bupranolol, carteolol, celiprolol, chlorpheniramine, cicloprolol, cimetidine, cinnarizine, cirazoline, clonidine, dilevalol, dimethindene, diphenhydramine, doxazosin, esmolol, famotidine, isothipendyl, ketotifen, metiamide, metoprolol, moxonidine, nadolol, naphazoline, nifenalol, nizatidine, oxprenolol, pheniramine, phentolamine, pindolol, pizotyline (pizotifen), practolol, prazosin, promethazine, propranolol, pyrilamine (mepyramine), ranitidine, roxatidine, sotalol, tiamenidine, timolol, tramazoline, tripeleminamine, triprolidine, tyamazoline, UK-14,304

REFERENCE

Kaliszan,R.; Nasal,A.; Turowski,M. Binding site for basic drugs on α₁-acid glycoprotein as revealed by chemometric analysis of biochromatographic data, *Biomed.Chromatogr.*, **1995**, 9, 211–215.

Chloroquine

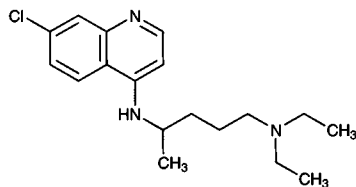
Molecular formula: C₁₈H₂₆ClN₃

Molecular weight: 319.88

CAS Registry No.: 54-05-7, 50-63-5 (phosphate), 3545-67-3 (HCl)

Merck Index: 2215

Lednicer No.: 1 341



SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 75 × 7.5 I.D. Progel-TSK Heparin-5PW affinity column (Supelco)

Mobile phase: 20 mM pH 6.0 sodium phosphate buffer

Column temperature: 19

Flow rate: 0.8

Detector: UV 214

CHROMATOGRAM

Retention time: 37 (+), 44 (-)

KEY WORDS

chiral

REFERENCE

Stalcup,A.M.; Gahm,K.H.; Baldueza,M. Chiral separation of chloroquine using heparin as a chiral selector in high-performance liquid chromatography, *Anal.Chem.*, **1996**, 68, 2248–2250.

SAMPLE

Matrix: blood

Sample preparation: 1 mL Plasma + 1 mL 1 M NaOH + 30 mL n-heptane, shake for 30 min. Remove 25 mL of the organic layer and evaporate it to dryness under a stream of nitrogen at 30°, reconstitute the residue in 100 µL MeOH:100 mM phosphoric acid 50:50, inject a 10 µL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 µm µBondapak C18

Mobile phase: MeCN:20 mM pH 3.4 1-heptanesulfonic acid 34:66 (Aqueous solution was 40 mL Pic B-7 (Waters) + 460 mL water, pH 3.4.)

Flow rate: 1

Injection volume: 10

Detector: UV 340

CHROMATOGRAM

Retention time: 9

Limit of detection: 5 ng

OTHER SUBSTANCES

Extracted: metabolites

KEY WORDS

plasma

REFERENCE

Brown,N.D.; Poon,B.T.; Chulay,J.D. Determination of chloroquine and its de-ethylated metabolites in human plasma by ion-pair high-performance liquid chromatography, *J.Chromatogr.*, **1982**, 229, 248–254.

SAMPLE

Matrix: blood

Sample preparation: 1 mL Whole blood or plasma + 1 mL water + 50 μ L 10 μ g/mL IS in MeOH + 1 mL 60% aqueous KOH, vortex for 1 min, heat for 3 min on a boiling water bath, pass onto a 3 mL Extrelut cartridge. Elute with diethyl ether:dichloromethane 70:30, evaporate eluate to dryness under a stream of air at 40°, vortex in 100 μ L initial mobile phase, inject a 40 μ L aliquot.

HPLC VARIABLES

Column: 300 \times 3.9 10 μ m μ Bondapak C18

Mobile phase: Gradient. A was KH_2PO_4 : 1 N phosphoric acid 999:1. B was MeCN. A:B 65:35 for 12 min at 1 mL/min and UV 343 nm then 55:45 at 2 mL/min and UV 242 nm (step gradient).

Flow rate: 1-2

Injection volume: 40

Detector: UV 343, UV 242

CHROMATOGRAM

Retention time: 5.35

Internal standard: papaverine hydrochloride (10.7 min)

Limit of detection: 10 ng/mL

OTHER SUBSTANCES

Extracted: metabolites, diazepam, monodesethylchloroquine, nordiazepam

Simultaneous: epinephrine, sulfadoxine, quinine, pyrimethamine, mefloquine

KEY WORDS

whole blood; plasma

REFERENCE

Estadieu,M.; Durand,A.; Viala,A.; Rop,P.P.; Fornaris,M.; Quicke,J. A rapid HPLC procedure for the simultaneous determination of chloroquine, monodesethylchloroquine, diazepam, and nordiazepam in blood, *J.Anal.Toxicol.*, **1989**, 13, 89–93.

SAMPLE

Matrix: blood

Sample preparation: Serum. 200 μ L Serum + 50 μ L 1 μ g/mL IS in water + 50 μ L 4 M NaOH + 200 μ L MTBE, vortex for 30 s, centrifuge at 9950 g for 2 min, inject 100 μ L of the organic layer. Whole blood. 100 μ L Whole blood + 500 μ L water + 50 μ L 1 μ g/mL IS in water + 50 μ L 4 M NaOH + 200 μ L MTBE, vortex for 30 s, centrifuge at 9950 g for 2 min, inject 100 μ L of the organic layer. Dried blood. Spread 100 μ L whole blood on a 70 \times 30 mm piece of filter paper, allow to dry, cut paper into 10 \times 5 mm strips, add 100 μ L 1 μ g/mL IS in water, add 1.5 mL 0.5 M NaOH, vortex for 30 s, let stand for 30 min at room temperature, add 300 μ L MTBE, vortex for 30 s, centrifuge at 2000 g for 5 min, inject a 100 μ L aliquot of the organic layer.

HPLC VARIABLES

Column: 150 \times 5 5 μ m Spherisorb S5SCX sulfophenylpropyl-modified silica

Mobile phase: MeOH:water 98.5:1.5 containing 9.41 g/L ammonium perchlorate, adjust apparent pH to 8.0 with 220 mL/L 50 mM NaOH in MeOH

Flow rate: 1.5

Injection volume: 100

Detector: F ex 215 em no filter

CHROMATOGRAM

Retention time: 14

Internal standard: 6,8-dichloro-4-(1-methyl-4-diethylaminobutylamino)quinoline (5)

Limit of quantitation: 5 ng/mL (serum), 10 ng/mL (whole blood, dried blood)

OTHER SUBSTANCES

Extracted: hydroxychloroquine, quinine, metabolites

Simultaneous: acebutolol, N-acetylprocainamide, atenolol, butriptyline, chlorpromazine, desipramine, flecainide, fluoxetine, imipramine, labetalol, maprotiline, mepacrine, metoprolol, mexiletine, norbutriptyline, normaprotiline, procainamide, propranolol, sotalol

Noninterfering: amitriptyline, amodiaquin, carbamazepine, clomipramine, dapsone, diazepam, dothiepin, doxepin, fluvoxamine, lorazepam, mefloquine, nitrazepam, norclomipramine, nordiazepam, nordothiepin, nordoxepin, nortriptyline, primaquine, proguanil, pyrimethamine

KEY WORDS

serum; whole blood; dried blood

REFERENCE

Croes, K.; McCarthy, P.T.; Flanagan, R.J. Simple and rapid HPLC of quinine, hydroxychloroquine, chloroquine, and desethylchloroquine in serum, whole blood, and filter paper-adsorbed dry blood, *J. Anal. Toxicol.*, **1994**, *18*, 255–260.

SAMPLE

Matrix: blood

Sample preparation: 2 mL Whole blood or plasma + 2 mL buffer + 5 mL chloroform: isopropanol:n-heptane 60:14:26, shake gently horizontally for 10 min, centrifuge at 2800 g for 10 min. Remove the lower organic layer and evaporate it to dryness under vacuum at 45°, reconstitute the residue in 100 µL mobile phase, centrifuge at 2800 g for 5 min, inject a 50 µL aliquot of the supernatant. (Buffer was saturated ammonium chloride solution 25% diluted with water, adjusted to pH 9.5 with 25% ammonia solution.)

HPLC VARIABLES

Column: 300 × 3.9 4 µm NovaPack C18

Mobile phase: MeOH:THF:buffer 65:5:30 (Buffer was 0.68 g/L (10 mM (sic)) KH₂PO₄ adjusted to pH 2.6 with concentrated orthophosphoric acid.) (At the end of each session wash the column with water for 1 h and MeOH for 1 h, re-equilibrate for 30 min.)

Column temperature: 30

Flow rate: 0.8

Injection volume: 50

Detector: UV 222

CHROMATOGRAM

Retention time: 4.72

Limit of detection: <120 ng/mL

KEY WORDS

whole blood; plasma; interferences may occur—compounds(all of which are extracted) elute in this order tenoxicam; iproniazid; methocarbamol; methotrexate; caffeine; nialamide; colchicine; cytarabine; benzoylecgonine; acetaminophen; diazoxide; dacarbazine; sulfinpyrazole; flumazenil; sulpride; morphine; atenolol; toloxatone; terbutaline; albuterol; phenobarbital; ranitidine; tiapride; phenol; chlormezanone; aspirin; metformin; ritodrine; codeine; sultopride; amisulpride; naltrexone; lisinopril; benzocaine; nizatidine; nalorphine; mephenesin; naloxone; sotalol; carteolol; procainamide; carbamazepine; bromazepam; nalbuphine; nadolol; procarbazine; dihydralazine; omeprazole; strychnine; acebutolol; glutethimide; chlorpropamide; glipizide; triazolam; prazosin; flunitrazepam;

clonazepam; metoclopramide; melphalan; estazolam; tolbutamide; ephedrine; clonidine; pindolol; clobazam; minoxidil; disopyramide; nitrazepam; dextromethorphan; tofisopam; zopiclone; debrisoquine; sulindac; alprazolam; cycloguanil; lorazepam; methaqualone; ketamine; piroxicam; metoprolol; nifedipine; quinine; mephentermine; prilocaine; pentazocine; oxazepam; tiaprofenic acid; quinidine; celiprolol; ajmaline; yohimbine; lidocaine; secobarbital; viloxazine; mepivacaine; meperidine; doxylamine; labetalol; temazepam; amodiaquine; benperidol; droperidol; hydroxychloroquine; zolpidem; ketoprofen; alminoprofen; cicletanine; moclobemide; chloroquine; cocaine; timolol; nomifensine; ticlopidine; acenocoumarol; vindesine; mexiletine; dipyridamole; trazodone; pipamperone; pyrimethamine; benazepril; vincristine; metapramine; chlordiazepoxide; oxprenolol; warfarin; clorazepate; flecainide; phenacyclidine; thiopental; fenfluramine; metipranolol; triprolidine; naproxen; buprenorphine; verapamil; buspirone; tianeptine; midazolam; bupivacaine; carbinoxamine; loperazole; cetirizine; chlorpheniramine; moperone; cibenzoline; medifoxamine; astemizole; vinblastine; nicardipine; bisoprolol; diltiazem; glibornuride; reserpine; aconitine; nitrendipine; diazepam; mianserin; ramipril; haloperidol; tetracaine; alprenolol; aceprometazine; glibenclamide; chlorophenacinone; doxepin; nimodipine; diphenhydramine; cyclizine; histapyrrodine; phenylbutazone; demexiptiline; clozapine; proguanil; trifluoperidol; medazepam; cyamemazine; bumadizone; suriclone; propranolol; acepromazine; dothiepin; dextromoramide; fenoprofen; dextropropoxyphene; loxapine; betaxolol; propafenone; promethazine; thioproperazine; methadone; amoxapine; quinupramine; opipramol; cyproheptadine; brompheniramine; mefenidramine; protriptyline; flurbiprofen; tetrazepam; zorubicin; prazepam; alimemazine; loperamide; imipramine; desipramine; levomepromazine; hydroxyzine; niflumic acid; penbutolol; fluvoxamine; pimozone; daunorubicin; indomethacin; maprotiline; tropatenine; etodolac; fluoxetine; amitriptyline; nortriptyline; tiocloamarol; diclofenac; mefloquine; trimipramine; chlorambucil; lidoflazine; ibuprofen; floctafenine; alpidem; loratadine; chlorpromazine; clomipramine; carpipramine; thioridazine; fentiazac; clemastine; mefenamic acid; fluphenazine; prochlorperazine; penfluridol; bepridil; terfenadine; trifluoperazine

REFERENCE

Tracqui,A.; Kintz,P.; Mangin,P. Systematic toxicological analysis using HPLC/DAD, *J.Forensic Sci.*, **1995**, *40*, 254-262.

SAMPLE

Matrix: blood

Sample preparation: 1 mL Serum + 100 μ L water containing 5 μ g/mL 2,3-diaminonaphthalene and 3.5 μ g/mL 18-hydroxy-11-deoxycorticosterone + 1 mL 250 mM NaOH + 7 mL diethyl ether, shake on a rotary shaker for 15 min, repeat extraction. Combine the organic layers and evaporate them to dryness under a stream of nitrogen at 30-40°, reconstitute the residue in 70 μ L MeOH:100 mM perchloric acid 50:50, inject a 20 μ L aliquot.

HPLC VARIABLES

Column: 150 \times 3.9 4 μ m Nova-Pak C18

Mobile phase: Gradient. A was 58 mM NaH₂PO₄ containing 6 mM sodium heptanesulfonate, adjusted to pH 3.1 with concentrated phosphoric acid. B was MeCN:MeOH 85:15. A: B from 100:0 to 78:22 over 5 min, to 70:30 over 12 min, maintain at 70:30 for 4 min, to 65:35 over 9 min.

Flow rate: 1

Injection volume: 20

Detector: UV 245, 256, 343

CHROMATOGRAM

Retention time: 13.20

Internal standard: 2,3-diaminonaphthalene (10.71), 18-hydroxy-11-deoxycorticosterone (15.85)

Limit of detection: 2 ng/mL (343 nm)

OTHER SUBSTANCES

Extracted: betamethasone, corticosterone, cortisone, dexamethasone, fluocinolone acetate, fluendrenolide, fluorometholone, fluprednisolone, hydrocortisone, hydroxychloroquine, 17 β -hydroxyprogesterone, meprednisone, methylprednisolone, methylprednisolone acetate, paramethasone, prednisolone, prednisone, progesterone, triamcinolone

Noninterfering: aspirin, ibuprofen, indomethacin, phenylbutazone, pregnenolone

KEY WORDS

serum

REFERENCE

Volin, P. Simple and specific reversed-phase liquid chromatographic, *J. Chromatogr. B*, **1995**, 666, 347–353.

SAMPLE

Matrix: blood

Sample preparation: Dilute urine 10-fold with water. 1 mL Plasma or urine + 4 mL diethyl ether + 10 (plasma) or 20 (urine) μ L 5 μ g/mL papaverine + 1 mL 2 M NaOH, vortex for 1 min, centrifuge at 2000 g for 10 min. Remove the organic layer and add it to 100 μ L 100 mM HCl, vortex for 1 min, inject a 10 μ L aliquot of the aqueous layer.

HPLC VARIABLES

Column: 300 \times 3.9 10 μ m Bondapak

Mobile phase: MeCN:MeOH:200 mM NaH₂PO₄ 5:30:65 containing 10 mL/L perchloric acid, pH 3.0

Flow rate: 1

Injection volume: 10

Detector: UV 254

CHROMATOGRAM

Retention time: 4

Internal standard: papaverine (6)

Limit of detection: 3 ng/mL

OTHER SUBSTANCES

Extracted: metabolites

Noninterfering: halofantrine, mefloquine, pyrimethamine, sulfadoxine, tetracycline

KEY WORDS

plasma; pharmacokinetics

REFERENCE

Walker, O.; Ademowo, O.G. A rapid, cost-effective liquid chromatographic method for the determination of chloroquine and desethylchloroquine in biological fluids, *Ther. Drug Monit.*, **1996**, 18, 92–96.

SAMPLE

Matrix: blood, urine

Sample preparation: 100 μ L Urine or 1 mL plasma + 100 ng IS + 1 mL MeCN, centrifuge. Remove supernatant and add it to 1 mL buffer, add 6 mL dichloromethane:petroleum ether:isopropanol 45:45:10, rotate for 10 min. Remove the upper organic layer and evaporate it under nitrogen at 60°. Dissolve residue in 100 μ L mobile phase, inject. (Buffer contained 80 g NaHCO₃ and 30 g K₂CO₃ per liter, pH 9.5.)

HPLC VARIABLES

Column: 466 \times 5 5 μ m Spherisorb ODS

Mobile phase: MeCN:MeOH:45 mM pH 4.5 KH₂PO₄ 40:3:57, containing 40 g/L NaClO₄ and 40 g/L trimethylammonium chloride

Flow rate: 1
Injection volume: 100
Detector: UV 338

CHROMATOGRAM

Retention time: 6.5
Internal standard: 4-(3-dimethylaminopropyl)-4-chloroquinoline (5)
Limit of quantitation: < 10 ng/mL

OTHER SUBSTANCES

Simultaneous: metronidazole

KEY WORDS

plasma

REFERENCE

Okonkwo,P.O.; Eta,E.I. Simultaneous determination of chloroquine and metronidazole in human biological fluid by high-pressure liquid chromatography, *Life Sci.*, **1988**, 42, 539–545.

SAMPLE

Matrix: blood, urine

Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 µL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) µL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18

Column: 250 × 4.6 5 µm Symmetry C8 (Waters)

Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A: B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.

Column temperature: 30

Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)

Injection volume: 10-30

Detector: UV 221.6

CHROMATOGRAM

Retention time: 5.442

KEY WORDS

whole blood

REFERENCE

Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, 763, 149–163.

SAMPLE

Matrix: bulk, erythrocytes, urine

Sample preparation: Condition a 3 mL Bond Elut C8 SPE cartridge with 2 mL MeOH and 2 mL buffer. Hemolyze erythrocytes in water 1:3. Dilute urine with water 1:99. 1 mL Plasma, hemolyzed erythrocytes, or diluted urine + 100 μ L 5 μ g/mL hydroxychloroquine sulfate in MeOH:water 50:50, mix, add to the SPE cartridge, wash with 4 mL buffer, wash with 2 mL MeOH:buffer 50:50, elute with 3 mL MeOH:ammonia 99:1. Evaporate the eluate to dryness under a stream of nitrogen at 30°, reconstitute the residue in the initial mobile phase, vortex, inject a 50 μ L aliquot. (Prepare buffer by mixing equal volumes of 100 mM ammonium formate and 100 mM ammonia solution, pH 9.2.)

HPLC VARIABLES

Guard column: 10 \times 4 Inertsil

Column: 250 \times 4.5 μ m Inertsil

Mobile phase: Gradient. A was MeCN. B was MeOH:25% ammonia solution 92.5:7.5. A:B 78:22 for 3 min, then to 65:35 over 2 min (Waters curve no. 3), maintain at 65:35 for 20 min, return to 78:22 over 5 min (Waters curve no. 3).

Flow rate: 0.85

Injection volume: 50

Detector: F ex 325 em 375

CHROMATOGRAM

Retention time: 15.6

Internal standard: hydroxychloroquine sulfate (11.5)

Limit of detection: 4.7 ng/mL

Limit of quantitation: 5.6 ng/mL

OTHER SUBSTANCES

Extracted: quinine, monodesethylchloroquine, bidesethylchloroquine

Simultaneous: halofantrine, quinidine

Noninterfering: proguanil, cycloguanil, 4-chlorophenylbiguanide, amodiaquine, mefloquine, pyrimethamine, sulfadoxine, cinchonine, cinchonidine

KEY WORDS

plasma; SPE

REFERENCE

Chaulet, J.-F.; Robet, Y.; Prevosto, J.-M.; Soares, O.; Brazier, J.-L. Simultaneous determination of chloroquine and quinine in human biological fluids by high-performance liquid chromatography, *J. Chromatogr.*, **1993**, 613, 303–310.

SAMPLE

Matrix: formulations

Sample preparation: Dilute with saline, inject a 10 μ L aliquot.

HPLC VARIABLES

Column: 250 \times 4.6 Lichrosorb 10 RP 8

Mobile phase: THF:triethylamine:water 4:0.75:96, adjusted to a pH of 2.3 with 2 M nitric acid

Flow rate: 2

Injection volume: 10

Detector: UV 254

CHROMATOGRAM

Retention time: 5.2

KEY WORDS

injections; saline

REFERENCE

Martens, H.J.; de Goede, P.N.; van Loenen, A.C. Sorption of various drugs in polyvinyl chloride, glass, and polyethylene-lined infusion containers, *Am.J.Hosp.Pharm.*, **1990**, *47*, 369–373.

SAMPLE

Matrix: microsomal incubations

Sample preparation: Mix 200 μ L microsomal incubation with 200 μ L ice-cold MeCN, vortex for 1 min, keep at 4° for 10–15 min. Centrifuge at 600 g at 4° for 10 min, inject a 15 μ L aliquot of the supernatant.

HPLC VARIABLES

Guard column: 5 μ m Hypersil C18

Column: 150 \times 4.6 5 μ m Spherisorb C1

Mobile phase: MeOH:water 70:30 containing 7 mM (0.1%) triethylamine

Flow rate: 1

Injection volume: 15

Detector: F ex 250 em 380

CHROMATOGRAM

Retention time: 12.8–13.1

Limit of detection: 1 nM

Limit of quantitation: 78 nM

OTHER SUBSTANCES

Extracted: metabolites

Noninterfering: coumarin, diethyldithiocarbamate, quinidine, ketoconazole, mephenytoin, α -naphthoflavone, sulfaphenazole, tolbutamide

KEY WORDS

liver

REFERENCE

Ducharme, J.; Farinotti, R. Rapid and simple method to determine chloroquine and its desethylated metabolites in human microsomes by high-performance liquid chromatography with fluorescence detection, *J.Chromatogr.B*, **1997**, *698*, 243–250.

SAMPLE

Matrix: solutions

Sample preparation: Dissolve in MeOH:water 1:1 at a concentration of 50 μ g/mL, inject a 10 μ L aliquot.

HPLC VARIABLES

Column: 300 \times 3.9 10 μ m μ Bondapak C18

Mobile phase: MeOH:acetic acid:triethylamine:water 30:1.5:0.5:68

Flow rate: 1.5

Injection volume: 10

Detector: UV

CHROMATOGRAM

Retention time: k' 0.99

REFERENCE

Roos, R.W.; Lau-Cam, C.A. General reversed-phase high-performance liquid chromatographic method for the separation of drugs using triethylamine as a competing base, *J.Chromatogr.*, **1986**, *370*, 403–418.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 cellulose tris(4-tert-butylphenylcarbamate)

Mobile phase: Hexane:isopropanol 90:10

Flow rate: 0.5

Detector: UV

CHROMATOGRAM

Retention time: 30 (-), 34 (+)

KEY WORDS

chiral

REFERENCE

Okamoto, Y.; Aburatani, R.; Hatano, K.; Hatada, K. Optical resolution of racemic drugs by chiral HPLC on cellulose and amylose tris(phenylcarbamate) derivatives, *J. Liq. Chromatogr.*, **1988**, *11*, 2147–2163.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Zorbax RX

Mobile phase: Gradient. A was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 1 L water. B was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 200 mL water, make up to 1 L with MeCN. A:B from 100:0 to 0:100 over 30 min, maintain at 0:100 for 5 min.

Column temperature: 30

Flow rate: 2

Detector: UV 210

OTHER SUBSTANCES

Also analyzed: acepromazine, acetaminophen, acetophenazine, albuterol, aminophylline, amitriptyline, amobarbital, amoxapine, amphetamine, amylocaine, antipyrine, aprobarbital, aspirin, atenolol, atropine, avermectin, barbital, benzocaine, benzoic acid, benzotroponine, benzphetamine, berberine, bibucaine, bromazepan, brompheniramine, buprenorphine, buspirone, butabarbital, butacaine, butethal, caffeine, carbamazepine, carbromal, chloramphenicol, chlorothiazide, chloroxylonol, chlorphenesin, chlorpheniramine, chlorpromazine, chlorpropamide, chlortetracycline, cimetidine, cinchonidine, cinchonine, clenbuterol, clonazepam, clonixin, clorazepate, cocaine, codeine, colchicine, cortisolone, coumarin, cyclazoxime, cyclobenzaprine, cyclothiazide, cyheptamide, cymarin, danazol, danthron, dapsone, debrisoquine, desipramine, dexamethasone, dextromethorphan, dextropropoxyphene, diamorphine, diazepam, diclofenac, diethylpropion, diethylstilbestrol, diflunisal, digitoxin, digoxin, diltiazem, diphenhydramine, diphenoxylate, diprenorphine, dipyrone, disulfiram, dopamine, doxapram, doxepin, dronabinol, ephedrine, epinephrine, epinine, estradiol, estriol, estrone, ethacrynic acid, ethosuximide, etonitazene, etorphine, eugenol, famotidine, fenbendazole, fencamfamine, fenopropfen, fenproporex, fentanyl, flubendazole, flufenamic acid, flunitrazepam, 5-fluorouracil, fluoxymesterone, fluphenazine, furosemide, gentisic acid, gitoxigenin, glipizide, glunixin, glutethimide, glybenclamide, guaiacol, halazepam, haloperidol, hydrochlorothiazide, hydrocodone, hydrocortisone, hydromorphone, hydroxyquinoline, ibogaine, ibuprofen, iminostilbene, imipramine, indomethacin, isocarboxystiril, isocarboxazid, isoniazid, isoproterenol, isoxsuprine, ivermectin, ketamine, ketoprofen, kynurenic acid, levorphanol, lidocaine, lorazepam, lormetazepam, loxapine, mazindol, mebendazole, meclizine, meclofenamic acid, medazepam, mefenamic acid, megestrol, mepacrine, meperidine, mephentermine, mephenytoin, mephesin, mephobarbital, mepivacaine, mescaline, mesoridazine, methadone, methamphetamine, methapy-

rilene, methaqualone, methazolamide, methocarbamol, methoxamine, methsuximide, methyl salicylate, methyl dopa, methyl dopamine, methylphenidate, methylprednisolone, methyltestosterone, methyprylon, metoprolol, mibolerone, morphine, nadolol, nalorphine, naloxone, naltrexone, naphazoline, naproxen, nefopam, niacinamide, nicotine, niacin, nifedipine, niflumic acid, nitrazepam, norepinephrine, nortriptyline, noscapine, nyldrin, oxazepam, oxycodone, oxymorphone, oxyphenbutazone, oxytetracycline, papaverine, pargyline, pemoline, pentazocine, pentobarbital, persantine, phenacetin, phenazocine, phenazopyridine, phencyclidine, phendimetrazine, phenelzine, pheniramine, phenobarbital, phenothiazine, phensuximide, phentermine, phenylbutazone, phenylephrine, phenylpropanolamine, piperocaine, prazepam, prednisolone, primidone, probenecid, progesterone, propiomazine, propranolol, propylparaben, pseudoephedrine, puromycin, pyrilamine, pyrihydione, quazepam, quinaldic acid, quinidine, quinine, ranitidine, recinnamine, reserpine, resorcinol, saccharin, albuterol, salicylamide, salicylic acid, scopolamine, scopolletin, secobarbital, strychnine, sulfacetamide, sulfadiazine, sulfadimethoxine, sulfaethidole, sulfamerazine, sulfamethazine, sulfamethoxazole, sulfanilamide, sulfapyridine, sulfasoxazole, sulindac, tamoxifen, temazepam, testosterone, tetracaine, tetracycline, tetramisole, thebaine, theobromine, theophylline, thiabendazole, thiamine, thiamylal, thiobarbituric acid, thioridazine, thiosalicylic acid, thiothixene, thymol, tolazamide, tolazoline, tobutamide, tolmetin, tranlycypromine, triamcinolone, tribenzylamine, trichloromethiazide, trifluoperazine, trihexyphenidyl, trimethoprim, tripeleennamine, triprolidine, tropacocaine, tyramine, verapamil, vincamine, warfarin, yohimbine, zoxazolamine

REFERENCE

Hill,D.W.; Kind,A.J. Reversed-phase solvent gradient HPLC retention indexes of drugs, *J.Anal.Toxicol.*, **1994**, *18*, 233–242.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Chirex 3014 (Phenomenex)

Mobile phase: Hexane:1,2-dichloroethane:EtOH/trifluoroacetic acid 50:35:15 (EtOH/trifluoroacetic acid was premixed 20:1.)

Flow rate: 0.7-1

Injection volume: 20

Detector: UV 346

KEY WORDS

chiral; $\alpha = 1.22$ for enantiomers

REFERENCE

Cleveland,T. Pirkle-concept chiral stationary phases for the HPLC separation of pharmaceutical racemates, *J.Liq.Chromatogr.*, **1995**, *18*, 649–671.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 150 × 5 Spherisorb S5SCX

Mobile phase: MeOH:water 98.5:1.5 containing 80 mM ammonium perchlorate, adjusted to pH 8.0 with 50 mM NaOH in MeOH

Flow rate: 1.5

Detector: F ex 215 no emission filter

CHROMATOGRAM

Retention time: 13.5

Internal standard: hydroxychloroquine (8)

OTHER SUBSTANCES

Simultaneous: hydroquinine, quinine

REFERENCE

Croes, K.; McCarthy, P.T.; Flanagan, R.J. HPLC of basic drugs and quaternary ammonium compounds on microparticulate strong cation-exchange materials using methanolic or aqueous methanol eluents containing an ionic modifier, *J. Chromatogr. A*, **1995**, 693, 289–306.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 5 µm Supelcosil LC-DP (A) or 250 × 4.5 µm LiChrospher 100 RP-8 (B)

Mobile phase: MeCN:0.025% phosphoric acid:buffer 25:10:5 (A) or 60:25:15 (B) (Buffer was 9 mL concentrated phosphoric acid and 10 mL triethylamine in 900 mL water, adjust pH to 3.4 with dilute phosphoric acid, make up to 1 L.)

Flow rate: 0.6

Injection volume: 25

Detector: UV 229

CHROMATOGRAM

Retention time: 12.70 (A), 3.55 (B)

OTHER SUBSTANCES

Also analyzed: acebutolol, acepromazine, acetaminophen, acetazolamide, acetophenazine, albuterol, alprazolam, amitriptyline, amobarbital, amoxapine, antipyrine, atenolol, atropine, azatadine, baclofen, benzocaine, bromocriptine, brompheniramine, brotizolam, bupivacaine, buspirone, butabarbital, butalbital, caffeine, carbamazepine, cetirizine, chlorcyclizine, chlordiazepoxide, chlormezanone, chlorpheniramine, chlorpromazine, chlorpropamide, chlorprothixene, chlorthalidone, chlorzoxazone, cimetidine, cisapride, clomipramine, clonazepam, clonidine, clozapine, cocaine, codeine, colchicine, cyclizine, cyclobenzaprine, dantrolene, desipramine, diazepam, diclofenac, diflunisal, diltiazem, diphenhydramine, diphenidol, diphenoxylate, dipyridamole, disopyramide, dobutamine, doxapram, doxepin, droperidol, encainide, ethidium bromide, ethopropazine, fenoprofen, fentanyl, flavoxate, fluoxetine, fluphenazine, flurazepam, flurbiprofen, fluvoxamine, furosemide, glutethimide, glyburide, guaifenesin, haloperidol, homatropine, hydralazine, hydrochlorothiazide, hydrocodone, hydromorphone, hydroxychloroquine, hydroxyzine, ibuprofen, imipramine, indomethacin, ketoconazole, ketoprofen, ketorolac, labetalol, levorphanol, lidocaine, loratadine, lorazepam, lovastatin, loxapine, mazindol, mefenamic acid, meperidine, mephénytoin, mepivacaine, mesoridazine, metaproterenol, methadone, methdilazine, methocarbamol, methotrexate, methotrimeprazine, methoxamine, methyl-dopa, methylphenidate, metoclopramide, metolazone, metoprolol, metronidazole, midazolam, moclobemide, morphine, nadolol, nalbuphine, naloxone, naphazoline, naproxen, nifedipine, nizatidine, norepinephrine, nortriptyline, oxazepam, oxycodone, oxymetazoline, paroxetine, pemoline, pentazocine, pentobarbital, pentoxifylline, perphenazine, pheniramine, phenobarbital, phenol, phenolphthalein, phentolamine, phenylbutazone, phenyltoloxamine, phenytoin, pimozide, pindolol, piroxicam, pramoxine, prazepam, prazosin, procabecid, procainamide, procaine, prochlorperazine, procyclidine, promazine, promethazine, propafenone, propantheline, propiomazine, propofol, propranolol, protriptyline, quazepam, quinidine, quinine, racemethorphan, ranitidine, remoxipride, risperidone, salicylic acid, scopolamine, secobarbital, sertraline, sotalol, spironolactone, sulfipyrazone, sulindac, temazepam, terbutaline, terfenadine, tetracaine, theophylline, thiethylperazine, thiopental, thioridazine, thiothixene, timolol, tocinide, tolbutamide, tolmetin, trazodone, triamterene, triazolam, trifluoperazine, trifluoromazine, trimeprazine, trimethoprim, trimipramine, verapamil, warfarin, xylometazoline, yohimbine, zopiclone

KEY WORDS

also details of plasma extraction

REFERENCE

Koves,E.M. Use of high-performance liquid chromatography-diode array detection in forensic toxicology, *J.Chromatogr.A*, **1995**, 692, 103–119.

SAMPLE

Matrix: urine

Sample preparation: 400 μ L Urine + 400 μ L 1 M NaOH + 30 mL n-heptane, shake for 30 min. Remove 25 mL of the organic layer and evaporate it to dryness under a stream of nitrogen at 30°, reconstitute the residue in 100 μ L MeOH:100 mM phosphoric acid 50:50, inject a 30 μ L aliquot.

HPLC VARIABLES

Column: 300 \times 7.8 10 μ m μ Bondapak C18

Mobile phase: MeCN:20 mM pH 3.4 1-heptanesulfonic acid 35:65 (Aqueous solution was 40 mL Pic B-7 (Waters) + 460 mL water, pH 3.4.)

Flow rate: 2

Injection volume: 30

Detector: UV 340

CHROMATOGRAM

Retention time: 18

Limit of detection: 2 ng

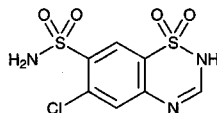
OTHER SUBSTANCES

Extracted: metabolites

REFERENCE

Brown,N.D.; Poon,B.T.; Chulay,J.D. Chloroquine metabolism in man: urinary excretion of 7-chloro-4-hydroxyquinoline and 7-chloro-4-aminoquinoline metabolites, *J.Chromatogr.*, **1985**, 345, 209–214.

Chlorothiazide



Molecular formula: $C_7H_6ClN_3O_4S_2$

Molecular weight: 295.73

CAS Registry No.: 58-94-6, 7085-44-1 (sodium salt)

Merck Index: 2221

Lednicer No.: 1 321

SAMPLE

Matrix: blood, CSF

Sample preparation: 200 μ L Serum, plasma, or CSF + 300 μ L reagent. Flush column A to waste with 500 μ L 500 mM ammonium sulfate, inject sample onto column A, flush column A to waste with 500 μ L 500 mM ammonium sulfate, elute the contents of column A onto column B with mobile phase, monitor the effluent from column B. (Reagent was 8.05 M guanidine hydrochloride and 1.02 M ammonium sulfate in water.)

HPLC VARIABLES

Column: A 30 \times 2.1 40 μ m preparative grade C18 (Analytichem); B 250 \times 4.6 10 μ m Partisil C8

Mobile phase: Gradient. A was 50 mM pH 4.5 KH_2PO_4 . B was MeCN:isopropanol 80:20. A: B 90:10 for 1 min, to 30:70 over 15 min, maintain at 30:70 for 4 min.

Column temperature: 50

Flow rate: 1.5

Detector: UV 280 for 5 min then UV 254

CHROMATOGRAM

Retention time: 5.43

Internal standard: heptanophenone (19.2)

OTHER SUBSTANCES

Extracted: acetazolamide, ampicillin, bromazepam, caffeine, carbamazepine, chloramphenicol, diazepam, droperidol, ethionamide, furosemide, isoniazid, methadone, penicillin G, phenobarbital, phenytoin, prazepam, propoxyphene, pyrazinamide, rifampin, trimeprazine, trimethoprim

KEY WORDS

plasma; serum; column-switching

REFERENCE

Seifart,H.I.; Kruger,P.B.; Parkin,D.P.; van Jaarsveld,P.P.; Donald,P.R. Therapeutic monitoring of anti-tuberculosis drugs by direct in-line extraction on a high-performance liquid chromatography system, *J.Chromatogr.*, **1993**, 619, 285-290.

SAMPLE

Matrix: blood, CSF, gastric contents, urine

Sample preparation: 200 μ L Serum, urine, CSF, or gastric fluid + 300 μ L reagent. Flush column A to waste with 500 μ L 500 mM ammonium sulfate, inject sample onto column A, flush column A to waste with 500 μ L 500 mM ammonium sulfate, backflush the contents of column A onto column B with mobile phase, monitor the effluent from column B. (Reagent was 8.05 M guanidine HCl and 1.02 M ammonium sulfate in water.)

HPLC VARIABLES

Column: A 40 μ m preparative grade C18 (Analytichem); B 75 \times 2.1 pellicular C18 (Whatman) + 250 \times 4.6 5 μ m C8 end-capped (Whatman)

Mobile phase: Gradient. A was 50 mM pH 4.5 KH_2PO_4 . B was MeCN:isopropanol 80:20. A: B 90:10 for 1 min, to 30:70 over 20 min.

Column temperature: 50

Flow rate: 1.5

Detector: UV 220

CHROMATOGRAM

Retention time: 5.67

Internal standard: heptanophenone (19)

OTHER SUBSTANCES

Extracted: acetaminophen, allobarbitol, azinphos, barbitol, brallobarbitone, bromazepam, butethal, caffeine, carbamazepine, carbaryl, cephaloridine, chloramphenicol, chlordiaze-poxide, chlorvinphos, clothiapine, cocaine, coomassie blue, desipramine, diazepam, di-phenhydramine, dipipanone, ethylbromphos, flufenamic acid, formothion, griseofulvin, in-domethacin, lidocaine, lorazepam, malathion, medazepam, midazolam, oxazepam, paraoxon, penicillin G, pentobarbital, prazepam, propoxyphene, prothiophos, quinine, sal-icylic acid, secobarbital, strychnine, sulfamethoxazole, theophylline, thiopental, thiorida-zine, trimethoprim

KEY WORDS

serum; column-switching

REFERENCE

Kruger,P.B.; Albrecht,C.F.De V.; Jaarsveld,P.P. Use of guanidine hydrochloride and ammonium sulfate in comprehensive in-line sorption enrichment of xenobiotics in biological fluids by high-performance liquid chromatography, *J.Chromatogr.*, **1993**, 612, 191–198.

SAMPLE

Matrix: formulations

Sample preparation: Grind tablet, add 10 mL DMSO, shake vigorously for 5 min, make up to 100 mL with MeOH, mix, filter (paper), discard the first 5 mL filtrate. Dilute 10 mL of the filtrate to 100 mL with MeOH, inject an aliquot.

HPLC VARIABLES

Column: 75 × 3.9 Novapak silica

Mobile phase: MeOH:20 g/L sodium 1-pentanesulfonate in water 100:1

Flow rate: 1

Injection volume: 20

Detector: UV 300

CHROMATOGRAM

Retention time: 5

OTHER SUBSTANCES

Simultaneous: reserpine (F ex 280 em 360)

KEY WORDS

tablets

REFERENCE

Cieri,U.R. Determination of reserpine and chlorothiazide in commercial tablets by liquid chromatogra-phy with fluorescence and UV absorbance detectors in series, *JAOAC Int.*, **1995**, 78, 1384–1387.

SAMPLE

Matrix: solutions

Sample preparation: Dilute 75 mL 1.2 mg/mL chlorothiazide in MeOH with 75 mL 380 mg/L IS in 0.1% phosphoric acid, inject an aliquot.

HPLC VARIABLES

Column: A 250 × 2 J sphere ODS-M80; B 150 × 4.6 5 µm Beckman Ultrasphere C18
Mobile phase: A Gradient. MeCN:0.1% formic acid from 0:100 to 30:70 over 20 min; B Gradient. MeCN:0.1% phosphoric acid from 0:100 to 30:70 over 12 min.
Flow rate: A 0.2; B 1
Detector: A MS, Finnigan Model TSQ-7000 triple-quadrupole, nebulizer nitrogen 260°; B UV 270

CHROMATOGRAM

Retention time: 20.5
Internal standard: ethylparaben

OTHER SUBSTANCES

Simultaneous: degradation products

KEY WORDS

photolysis

REFERENCE

Revelle,L.K.; Musser,S.M.; Rowe,B.J.; Feldman,I.C. Identification of chlorothiazide and hydrochlorothiazide UV-A photolytic decomposition products, *J.Pharm.Sci.*, **1997**, 86, 631–634.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 2 C18 glass lined (Whatman)
Mobile phase: MeCN:water 60:40
Flow rate: 0.04
Injection volume: 0.5
Detector: UV 254 or MS, Hewlett Packard 5985, home-made interface (details in paper)

CHROMATOGRAM

Retention time: 6.5

OTHER SUBSTANCES

Simultaneous: hydrochlorothiazide, trichlormethiazide

KEY WORDS

microbore

REFERENCE

Eckers,C.; Skrabalak,D.S.; Henion,J. On-line direct liquid introduction interface for micro-liquid chromatography/mass spectrometry: application to drug analysis, *Clin.Chem.*, **1982**, 28, 1882–1886.

SAMPLE

Matrix: solutions
Sample preparation: Dissolve in MeOH:water 1:1 at a concentration of 50 µg/mL, inject a 10 µL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 µm µBondapak C18
Mobile phase: MeOH:acetic acid:triethylamine:water 30:1.5:0.5:68
Flow rate: 1.5

Injection volume: 10

Detector: UV

CHROMATOGRAM

Retention time: k' 0.31

REFERENCE

Roos,R.W.; Lau-Cam,C.A. General reversed-phase high-performance liquid chromatographic method for the separation of drugs using triethylamine as a competing base, *J.Chromatogr.*, **1986**, 370, 403–418.

SAMPLE

Matrix: solutions

Sample preparation: Irradiate an ethanolic solution at 313 nm, inject a 3 µL aliquot.

HPLC VARIABLES

Column: 125 × 4.5 µm LiChrospher RP-18

Mobile phase: MeOH:water:acetic acid 10:88:2, pH 2.7

Flow rate: 1

Injection volume: 3

Detector: UV 265

CHROMATOGRAM

Retention time: 3.3

OTHER SUBSTANCES

Simultaneous: degradation products

REFERENCE

Ulvi,V.; Tammilehto,S. High-performance liquid chromatographic method for studies on the photodecomposition kinetics of chlorothiazide, *J.Chromatogr.*, **1990**, 507, 151–156.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Zorbax RX

Mobile phase: Gradient. A was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 1 L water. B was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 200 mL water, make up to 1 L with MeCN. A:B from 100:0 to 0:100 over 30 min, maintain at 0:100 for 5 min.

Column temperature: 30

Flow rate: 2

Detector: UV 210

OTHER SUBSTANCES

Also analyzed: acepromazine, acetaminophen, acetophenazine, albuterol, aminophylline, amitriptyline, amobarbital, amoxapine, amphetamine, amylocaine, antipyrine, aprobarbital, aspirin, atenolol, atropine, avermectin, barbital, benzocaine, benzoic acid, benzotropine, benzphetamine, berberine, bibucaine, bromazepan, brompheniramine, buprenorphine, buspirone, butabarbital, butacaine, butethal, caffeine, carbamazepine, carbromal, chloramphenicol, chlordiazepoxide, chloroxylenol, chlorphenesin, chlorpheniramine, chlorpromazine, chlorpropamide, chlortetracycline, cimetidine, cinchonidine, cinchonine, clenbuterol, clonazepam, clonixin, clorazepate, cocaine, codeine, colchicine, cortisone, coumarin, cyclazocine, cyclobenzaprine, cyclothiazide, cyheptamide, cymarin, danazol, danthron, dapsone, debrisoquine, desipramine, dexamethasone, dextromethorphan, dextropropoxyphene, diamorphine, diazepam, diclofenac, diethylpropion, diethylstilbestrol, diflunisal,

digitoxin, digoxin, diltiazem, diphenhydramine, diphenoxylate, diprenorphine, dipyrone, disulfiram, dopamine, doxapram, doxepin, dronabinol, ephedrine, epinephrine, epinine, estradiol, estriol, estrone, ethacrynic acid, ethosuximide, etonitazene, etorphine, eugenol, famotidine, fenbendazole, fencamfamine, fenpropfen, fenproporex, fentanyl, flubendazole, flufenamic acid, flunitrazepam, 5-fluorouracil, fluoxymesterone, fluphenazine, furosemide, gentisic acid, gitoxigenin, glipizide, glunixin, glutethimide, glybenclamide, guaiacol, halazepam, haloperidol, hydrochlorothiazide, hydrocodone, hydrocortisone, hydromorphone, hydroxyquinoline, ibogaine, ibuprofen, iminostilbene, imipramine, indomethacin, isocarboxtyril, isocarboxazid, isoniazid, isoproterenol, isoxsuprine, ivermectin, ketamine, ketoprofen, kynurenic acid, levorphanol, lidocaine, lorazepam, lormetazepam, loxapine, mazindol, mebendazole, meclizine, meclofenamic acid, medazepam, mefenamic acid, megestrol, mepacrine, meperidine, mephentermine, mephenytoin, mephesin, mephobarbital, mepivacaine, mescaline, mesoridazine, methadone, methamphetamine, methapyrilene, methaqualone, methazolamide, methocarbamol, methoxamine, methsuximide, methyl salicylate, methyl dopa, methyl dopamine, methylphenidate, methylprednisolone, methyltestosterone, methypylon, metoprolol, mibolerone, morphine, nadolol, nalorphine, naloxone, naltrexone, naphazoline, naproxen, nefopam, niacinamide, nicotine, niacin, nifedipine, niflumic acid, nitrazepam, norepinephrine, nortriptyline, noscapine, nyldrin, oxazepam, oxycodone, oxymorphone, oxyphenbutazone, oxytetracycline, papaverine, pargyline, pemoline, pentazocine, pentobarbital, persantine, phenacetin, phenazocine, phenazopyridine, phencyclidine, phendimetrazine, phenelzine, pheniramine, phenobarbital, phenothiazine, phensuximide, phentermine, phenylbutazone, phenylephrine, phenylpropanolamine, piperocaine, prazepam, prednisolone, primidone, probenecid, progesterone, propiomazine, propranolol, propylparaben, pseudoephedrine, puromycin, pyrilamine, pyrithyldione, quazepam, quinaldic acid, quinidine, quinine, ranitidine, recinnamine, reserpine, resorcinol, saccharin, albuterol, salicylamide, salicylic acid, scopolamine, scopoletin, secobarbital, strychnine, sulfacetamide, sufadiazine, sulfadimethoxine, sulfaethidole, sulfamerazine, sulfamethazine, sulfamethoxazole, sulfanilamide, sulfapyridine, sulfasoxazole, sulindac, tamoxifen, temazepam, testosterone, tetracaine, tetracycline, tetramisole, thebaine, theobromine, theophylline, thiabendazole, thiamine, thiamylal, thiobarbituric acid, thioridazine, thiosalicylic acid, thiothixene, thymol, tolazamide, tolazoline, tobutamide, tolmetin, tranlycypromine, triamcinolone, tribenzylamine, trichloromethiazide, trifluoperazine, trihexyphenidyl, trimethoprim, tripeleennamine, triprolidine, tropacocaine, tyramine, verapamil, vincamine, warfarin, yohimbine, zoxazolamine

REFERENCE

Hill,D.W.; Kind,A.J. Reversed-phase solvent gradient HPLC retention indexes of drugs, *J.Anal.Toxicol.*, **1994**, *18*, 233–242.

SAMPLE

Matrix: urine

Sample preparation: 2 mL Urine + 2 mL 1 M pH 4.1 NaH_2PO_4 + 4 mL ethyl acetate, vortex for 2 min, centrifuge at 1500 g for 5 min. Remove the organic phase and add it to 5 mL 100 mM pH 7.5 Na_2HPO_4 , vortex for 2 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 60°, reconstitute the residue in 100 μL MeCN:10 mM pH 3.0 phosphate buffer, inject a 5 μL aliquot.

HPLC VARIABLES

Column: 125 \times 4.5 μm LiCHrosorb RP-18

Mobile phase: Gradient. MeCN:10 mM pH 3.0 phosphate buffer 10:90 for 1.5 min then to 35:65 over 2 min

Column temperature: 50

Flow rate: 1.5

Injection volume: 5

Detector: UV 271

CHROMATOGRAM

Retention time: 1.8

Limit of quantitation: 1500 ng/mL

OTHER SUBSTANCES

Extracted: hydrochlorothiazide, quinethazone, chlorthalidone, methyclothiazide, clopamide, furosemide, metolazone, mefruside, bendroflumethiazide, cyclopenthiazide, bumetanide

Simultaneous: indapamide, clorexolone, ethacrynic acid

Noninterfering: aspirin, albuterol, allopurinol, alprenolol, atenolol, captopril, carbimazole, clonidine, coloxyl, danthron, diazepam, digoxin, doxepin, glibenclamide, hydralazine, indomethacin, labetalol, metformin, methyldopa, metoprolol, mianserin, minoxidil, nifedipine, nitrazepam, oxazepam, oxprenolol, pindolol, prazosin, propranolol, senokot, theophylline, trifluoperazine

REFERENCE

Fullinlaw, R.O.; Bury, R.W.; Moulds, R.F.W. Liquid chromatographic screening of diuretics in urine, *J.Chromatogr.*, **1987**, *415*, 347–356.

SAMPLE

Matrix: urine

Sample preparation: 2 mL Urine + 0.5 g solid buffer I (pH 5-5.5), vortex 15 s, add 4 mL ethyl acetate, agitate for 10 min, centrifuge at 600 g for 5 min. Remove organic layer and vortex it with 2 mL 5% aqueous lead acetate for 10 s, centrifuge at 600 g for 5 min, remove and keep organic phase. 2 mL Urine + 0.5 g solid buffer II (pH 9-9.5), vortex 15 s, add 4 mL ethyl acetate, agitate for 10 min, centrifuge at 600 g for 5 min. Remove organic layer and combine it with previous organic layer. Evaporate to dryness at 50° under a stream of nitrogen, reconstitute in 300 µL 50 µg/mL β-hydroxyethyltheophylline in MeOH, inject 5 µL aliquot. (Solid buffer I was $\text{KH}_2\text{PO}_4:\text{Na}_2\text{HPO}_4$ 99:1, solid buffer II was $\text{NaHCO}_3:\text{K}_2\text{CO}_3$ 3:2.)

HPLC VARIABLES

Column: 250 × 4.6 5 µm HP Hypersil ODS (A) or HP LiChrosorb RP-18 (B)

Mobile phase: Gradient. MeCN:buffer from 15:85 at 2 min to 80:20 at 20 min (Buffer was 50 mM NaH_2PO_4 containing 16 mM propylamine hydrochloride, adjusted to pH 3 with concentrated phosphoric acid.)

Flow rate: 1

Injection volume: 5

Detector: UV 230, UV 275

CHROMATOGRAM

Retention time: 5.37 (A), 6.43 (B)

Internal standard: β-hydroxyethyltheophylline (3.7 (A), 4.4 (B))

Limit of detection: 1000 ng/mL

OTHER SUBSTANCES

Extracted: furosemide, metolazone, amiloride, acetazolamide, hydrochlorothiazide, quinethazone, triamterene, flumethiazide, hydroflumethiazide, chlorthalidone, dichlorphenamide, trichloromethiazide, methyclothiazide, benzthiazide, cyclothiazide, polythiazide, bendroflumethiazide, ethacrynic acid, bumetanide, probenecid, spironolactone, canrenone

Noninterfering: acetaminophen, aspirin, caffeine, diflunisal, fenoprofen, ibuprofen, indomethacin, methocarbamol, naproxen, phenylbutazone, sulindac, tetracycline, theobromine, theophylline, tolmetin, trimethoprim, verapamil

REFERENCE

Cooper, S.F.; Massé, R.; Dugal, R. Comprehensive screening procedure for diuretics in urine by high-performance liquid chromatography, *J.Chromatogr.*, **1989**, *489*, 65–88.

SAMPLE

Matrix: urine

Sample preparation: Buffer urine to 4.9 by mixing with an equal volume of pH 4.9 200 mM sodium phosphate buffer. Inject a 40 μ L aliquot onto column A with mobile phase A, after 3 min backflush the contents of column A onto column B with mobile phase B and start the gradient. At the end of the run re-equilibrate for 10 min.

HPLC VARIABLES

Column: A 20 \times 4 5 μ m Hypersil octadecylsilica ODS; B 200 \times 4.6 5 μ m Shiseido SG-120 polymer-based C18

Mobile phase: A water; B Gradient. MeCN:buffer from 7:93 to 15:85 over 3.5 min, to 50:50 over 8.5 min, maintain at 50:50 for 11 min (Buffer was 6.9 g $\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$ in 1 L water, pH adjusted to 3.1 with phosphoric acid.)

Flow rate: 1

Injection volume: 40

Detector: UV 230

CHROMATOGRAM

Retention time: 10.8

Limit of detection: 1000 ng/mL

OTHER SUBSTANCES

Extracted: acetazolamide, amiloride, bendroflumethiazide, benzthiazide, bumetanide, caffeine, carbamazepine, chlorthalidone, clopamide, dichlorfenamide, ethacrynic acid, furosemide, hydrochlorothiazide, metyrapone, probenecid, spironolactone, triamterene, trichlormethiazide

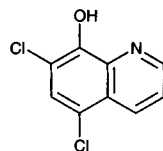
KEY WORDS

column-switching; optimum detection wavelengths vary for each drug

REFERENCE

Saarinén, M.; Sirén, H.; Riekkola, M.-L. A column switching technique for the screening of diuretics in urine by high performance liquid chromatography, *J. Liq. Chromatogr.*, **1993**, 16, 4063–4078.

Chloroxine



Molecular formula: C₉H₅Cl₂NO

Molecular weight: 214.05

CAS Registry No.: 773-76-2

Merck Index: 2227

SAMPLE

Matrix: formulations

Sample preparation: Weigh out shampoo containing 30 mg of chloroxine, add 40-50 mL MeOH, dissolve with heating, make up to 100 mL with MeOH. Remove a 2 mL aliquot and add it to 1 mL 10 mg/mL nickel chloride in MeOH, add 3 mL 0.4 mg/mL diphenylamine in MeOH, make up to 50 mL with MeOH, filter, inject an aliquot.

HPLC VARIABLES

Column: 300 × 3.9 μBondapak phenyl

Mobile phase: MeCN:MeOH:water 30:20:50 containing 1 mM NiCl₂

Flow rate: 1.2

Detector: UV 273

CHROMATOGRAM

Retention time: 6.5

Internal standard: diphenylamine (11)

OTHER SUBSTANCES

Simultaneous: iodochlorhydroxyquin, iodoquinol

KEY WORDS

shampoo; separated as Ni chelates

REFERENCE

Wojtowicz, E.J. Reverse-phase high-performance liquid chromatographic determination of halogenated 8-hydroxyquinoline compounds in pharmaceuticals and bulk drugs, *J.Pharm.Sci.*, **1984**, *73*, 1430-1433.

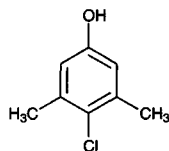
Chloroxylenol

Molecular formula: C₈H₉ClO

Molecular weight: 156.61

CAS Registry No.: 88-04-0

Merck Index: 2228



SAMPLE

Matrix: formulations

Sample preparation: Sonicate an amount of cream or ointment containing 230 µg chloroxylenol with 10 mL DMSO at 40° for 10 min, decant. Sonicate a 200 µL aliquot of the clear solution with 30 µL 60 mM potassium carbonate solution for 10 min. Add 400 µL 2.5 mg/mL 2-chloro-6,7-dimethoxy-3-quinoline carboxaldehyde solution in DMSO. Stir the reaction mixture at 110° for 50 min. Add 60 µL 2 M acetic acid and 800 µL IS in the mobile phase. Sonicate the mixture for 1 min. Inject a 50 µL aliquot. (Details for the preparation of 2-chloro-6,7-dimethoxy-3-quinoline carboxaldehyde are given in *Tetrahedron Letters* 1978, 23, 2045.)

HPLC VARIABLES

Column: 250 × 4.6 Hypersil 50DS

Mobile phase: MeCN:THF:50 mM pH 3.0 triethyl ammonium phosphate buffer 49.4:2.6:48

Column temperature: 35

Flow rate: 1.3

Injection volume: 50

Detector: F ex 360 em 500

CHROMATOGRAM

Retention time: 27.8 (4-chloro-3,5-xyleneol)

Internal standard: 4-chloro-3-cresol (19.5)

Limit of detection: 1 pmol

KEY WORDS

derivatization; cream; ointment

REFERENCE

Gatti,R.; Roveri,P.; Bonazzi,D.; Cavrini,V. HPLC-fluorescence determination of chlorocresol and chloroxylenol in pharmaceuticals, *J.Pharm.Biomed.Anal.*, **1997**, 16, 405–412.

SAMPLE

Matrix: solutions

Sample preparation: Prepare a 0.5 mg/mL solution in MeOH, inject a 5 µL aliquot.

HPLC VARIABLES

Column: 250 × 4.6 Zorbax RX

Mobile phase: Gradient. A was 150 mM phosphoric acid and 50 mM triethylamine. B was MeCN:water 80:20 containing 150 mM phosphoric acid and 50 mM triethylamine. A:B 100:0 for 2.2 min then to 0:100 over 30 min.

Column temperature: 30

Flow rate: 2

Injection volume: 5

Detector: UV 210

CHROMATOGRAM

Retention time: 21.4

OTHER SUBSTANCES

Simultaneous: acetaminophen, aprobarbital, butabarbital, chlordiazepoxide, chlorpromazine, clenbuterol, cortisone, danazol, diflunisal, doxapram, estrone, fluoxymesterone, mefenamic acid, methyltestosterone, nicotine, oxazepam, phentermine, phenylpropanolamine, progesterone, sulfamethazine, sulfanilamide, testosterone, testosterone propionate, tranlycypromine, tripeleennamine

KEY WORDS

details for purification of triethylamine in paper

REFERENCE

Hill,D.W.; Kind,A.J. The effects of type B silica and triethylamine on the retention of drugs in silica based reverse phase high performance chromatography, *J.Liq.Chromatogr.*, **1993**, *16*, 3941-3964.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Zorbax RX

Mobile phase: Gradient. A was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 1 L water. B was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 200 mL water, make up to 1 L with MeCN. A:B from 100:0 to 0:100 over 30 min, maintain at 0:100 for 5 min.

Column temperature: 30

Flow rate: 2

Detector: UV 210

OTHER SUBSTANCES

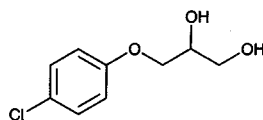
Also analyzed: acepromazine, acetaminophen, acetophenazine, albuterol, aminophylline, amitriptyline, amobarbital, amoxapine, amphetamine, amylocaine, antipyrine, aprobarbital, aspirin, atenolol, atropine, avermectin, barbital, benzocaine, benzoic acid, benzotropine, benzphetamine, berberine, bibucaine, bromazepan, brompheniramine, buprenorphine, buspirone, butabarbital, butacaine, butethal, caffeine, carbamazepine, carbomal, chloramphenicol, chlordiazepoxide, chloroquine, chlorphenesin, chlorpheniramine, chlorpromazine, chlorpropamide, chlortetracycline, cimetidine, cinchonidine, cinchonine, clenbuterol, clonazepam, clonixin, clorazepate, cocaine, codeine, colchicine, cortisone, coumarin, cyclazocine, cyclobenzaprine, cyclothiazide, cyheptamide, cymarin, danazol, danthron, dapsone, debrisoquine, desipramine, dexamethasone, dextromethorphan, dextropropoxyphene, diamorphine, diazepam, diclofenac, diethylpropion, diethylstilbestrol, diflunisal, digitoxin, digoxin, diltiazem, diphenhydramine, diphenoxylate, diprenorphine, dipyrone, disulfiram, dopamine, doxapram, doxepin, dronabinol, ephedrine, epinephrine, epinine, estradiol, estril, estrone, ethacrynic acid, ethosuximide, etonitazene, etorphine, eugenol, famotidine, fenbendazole, fencamfamine, fenoprofen, fenproporex, fentanyl, flubendazole, flufenamic acid, flunitrazepam, 5-fluorouracil, fluoxymesterone, fluphenazine, furosemide, gentisic acid, gitoxigenin, glipizide, glunixin, glutethimide, glybenclamide, guaiacol, halazepam, haloperidol, hydrochlorothiazide, hydrocodone, hydrocortisone, hydromorphone, hydroxyquinoline, ibogaine, ibuprofen, iminostilbene, imipramine, indomethacin, isocarboxtyril, isocarboxazid, isoniazid, isoproterenol, isoxsuprine, ivermectin, ketamine, ketoprofen, kynurenic acid, levorphanol, lidocaine, lorazepam, lormetazepam, loxapine, mazindol, mebendazole, meclizine, meclofenamic acid, medazepam, mefenamic acid, megestrol, mepacrine, meperidine, mephentermine, mephentyoin, mephesin, mephobarbital, mepivacaine, mescaline, mesoridazine, methadone, methamphetamine, methapyrilene, methaqualone, methazolamide, methocarbamol, methoxamine, methsuximide, methyl salicylate, methyl dopa, methyl dopamine, methylphenidate, methylprednisolone, methyltestosterone, methyprylon, metoprolol, mibolerone, morphine, nadolol, nalorphine, naloxone, naltrexone, naphazoline, naproxen, nefopam, niacinamide, nicotine, niacin, nifedipine, niflumic acid, nitrazepam, norepinephrine, nortriptyline, noscapine, nyldrin, oxazepam, oxycodone, oxymorphone, oxyphenbutazone, oxytetracycline, papaverine, pargy-

line, pemoline, pentazocine, pentobarbital, persantine, phenacetin, phenazocine, phenazopyridine, phencyclidine, phendimetrazine, phenelzine, pheniramine, phenobarbital, phenothiazine, phensuximide, phentermine, phenylbutazone, phenylephrine, phenylpropanolamine, piperocaine, prazepam, prednisolone, primidone, probenecid, progesterone, propiomazine, propranolol, propylparaben, pseudoephedrine, puromycin, pyrilamine, pyrithyldione, quazepam, quinaldic acid, quinidine, quinine, ranitidine, recinnamine, reserpine, resorcinol, saccharin, albuterol, salicylamide, salicylic acid, scopolamine, scopolletin, secobarbital, strychnine, sulfacetamide, sulfadiazine, sulfadimethoxine, sulfaethiodole, sulfamerazine, sulfamethazine, sulfamethoxazole, sulfanilamide, sulfapyridine, sulfasoxazole, sulindac, tamoxifen, temazepam, testosterone, tetracaine, tetracycline, tetramisole, thebaine, theobromine, theophylline, thiabendazole, thiamine, thiamylal, thiobarbituric acid, thioridazine, thiosalicylic acid, thiothixene, thymol, tolazamide, tolazoline, tobutamide, tolmetin, tranlycypromine, triamcinolone, tribenzylamine, trichloromethiazide, trifluoperazine, trihexyphenidyl, trimethoprim, tripeleennamine, triprolidine, tropacocaine, tyramine, verapamil, vincamine, warfarin, yohimbine, zoxazolamine

REFERENCE

- Hill,D.W.; Kind,A.J. Reversed-phase solvent gradient HPLC retention indexes of drugs, *J.Anal.Toxicol.*, **1994**, *18*, 233-242.

Chlorphenesin



Molecular formula: $C_9H_{11}ClO_2$

Molecular weight: 202.64

CAS Registry No.: 104-29-0

Merck Index: 2230

Lednicer No.: 1 118

SAMPLE

Matrix: reaction mixtures

Sample preparation: 5 mL Reaction mixture + 1 mL 5 M HCl, add 2 g NaCl, add 2 mL dichloromethane:n-butanol 50:50, shake for 5 min, centrifuge at 3000 rpm for 10 min. Remove the upper organic layer and extract the lower aqueous layer with 2 mL and with 1 mL of dichloromethane:n-butanol 50:50. Combine the organic layers and make up to 5 mL with dichloromethane:n-butanol 50:50, inject a 10 μ L aliquot.

HPLC VARIABLES

Column: 500 \times 4.5 μ m Lichrosorb SI 100

Mobile phase: Water-saturated dichloromethane:dichloromethane:n-butanol:n-hexane:MeOH 38.5:38.5:10:10:3

Flow rate: 1.3

Injection volume: 10

Detector: UV 233

CHROMATOGRAM

Retention time: 9

OTHER SUBSTANCES

Simultaneous: chlorphenesin carbamate

KEY WORDS

normal phase

REFERENCE

Hara,M.; Hayashi,H.; Yoshida,T.; Murayama,H. Studies on the kinetics and mechanism of drug degradation. I. Kinetics and mechanism of degradation of chlorphenesin carbamate in strongly alkaline aqueous solutions, *Chem.Pharm.Bull.(Tokyo)*, **1986**, 34, 1764–1769.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 125 \times 3.5 μ m Lichrospher 60 RP-Select-B

Mobile phase: B MeOH:30 mM ammonium formate 40:60

Flow rate: 0.5

Detector: UV 230

CHROMATOGRAM

Retention time: 7.2

OTHER SUBSTANCES

Simultaneous: cloxyquin, naftifine, sulbentine, tolnaftate, degradation products

REFERENCE

Thoma,K.; Kübler,N.; Reimann,E. Untersuchung der Photostabilität von Antimykotika. 3. Mitteilung: Photostabilität lokal wirksamer Antimykotika [Photodegradation of antimycotic drugs. 3. Communication: Photodegradation of topical antimycotics], *Pharmazie*, **1997**, *52*, 362–373.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 125 × 4.5 µm LiChrospher 60 RP-Select-B

Mobile phase: MeOH:20 mM ammonium formate buffer 40:60

Flow rate: 1

Detector: UV 230

CHROMATOGRAM

Retention time: 4.69

OTHER SUBSTANCES

Simultaneous: cloxyquin, naftifine, sulbentine, tolnaftate, degradation products

REFERENCE

Thoma,K.; Kübler,N.; Reimann,E. Untersuchung der Photostabilität von Antimykotika. 3. Mitteilung: Photostabilität lokal wirksamer Antimykotika [Photodegradation of antimycotic drugs. 3. Communication: Photodegradation of topical antimycotics], *Pharmazie*, **1997**, *52*, 362–373.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Zorbax RX

Mobile phase: Gradient. A was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 1 L water. B was 10 mL concentrated orthophosphoric acid and 7 mL triethylamine in 200 mL water, make up to 1 L with MeCN. A:B from 100:0 to 0:100 over 30 min, maintain at 0:100 for 5 min.

Column temperature: 30

Flow rate: 2

Detector: UV 210

OTHER SUBSTANCES

Also analyzed: acepromazine, acetaminophen, acetophenazine, albuterol, aminophylline, amitriptyline, amobarbital, amoxapine, amphetamine, amyllocaine, antipyrine, aprobarbital, aspirin, atenolol, atropine, avermectin, barbital, benzocaine, benzoic acid, benzotropine, benzphetamine, berberine, bibucaine, bromazepan, brompheniramine, buprenorphine, buspirone, butabarbital, butacaine, butethal, caffeine, carbamazepine, carbromal, chloramphenicol, chlordiazepoxide, chloroquine, chlorothiazide, chlorpheniramine, chlorpromazine, chlorpropamide, chlortetracycline, cimetidine, cinchonidine, cinchonine, clenbuterol, clonazepam, clonixin, clorazepate, cocaine, codeine, colchicine, cortisolone, coumarin, cyclazocine, cyclobenzaprine, cyclothiazide, cyheptamide, cymarin, danazol, danthron, dapson, debrisoquine, desipramine, dexamethasone, dextromethorphan, dextropropoxyphene, diamorphine, diazepam, diclofenac, diethylpropion, diethylstilbestrol, diflunisal, digitoxin, digoxin, diltiazem, diphenhydramine, diphenoxylate, diprenorphine, dipyrone, disulfiram, dopamine, doxapram, doxepin, dronabinol, ephedrine, epinephrine, epinine, estradiol, estriol, estrone, ethacrynic acid, ethosuximide, etonitazene, etorphine, eugenol, famotidine, fenbendazole, fencamfamine, fenopropfen, fenproporex, fentanyl, flubendazole, flufenamic acid, flunitrazepam, 5-fluorouracil, fluoxymesterone, fluphenazine, furosemide, gentisic acid, gitoxigenin, glipizide, glunixin, glutethimide, glybenclamide, guaiacol, halazepam, haloperidol, hydrochlorothiazide, hydrocodone, hydrocortisone, hydromorphone,

hydroxyquinoline, ibogaine, ibuprofen, iminostilbene, imipramine, indomethacin, isocarboxtyril, isocarboxazid, isoniazid, isoproterenol, isoxsuprine, ivermectin, ketamine, ketoprofen, kynurenic acid, levorphanol, lidocaine, lorazepam, lormetazepam, loxapine, mazindol, mebendazole, meclizine, meclofenamic acid, medazepam, mefenamic acid, megestrol, mepacrine, meperidine, mephentermine, mephenytoin, mephesin, mephobarbital, mepivacaine, mescaline, mesoridazine, methadone, methamphetamine, methapyrilene, methaqualone, methazolamide, methocarbamol, methoxamine, methsuximide, methyl salicylate, methyl dopa, methyl dopamine, methylphenidate, methylprednisolone, methyltestosterone, methyprylon, metoprolol, mibolerone, morphine, nadolol, nalorphine, naloxone, naltrexone, naphazoline, naproxen, nefopam, niacinamide, nicotine, niacin, nifedipine, niflumic acid, nitrazepam, norepinephrine, nortriptyline, noscapine, nyldrin, oxazepam, oxycodone, oxymorphone, oxyphenbutazone, oxytetracycline, papaverine, pargyline, pemoline, pentazocine, pentobarbital, persantine, phenacetin, phenazocine, phenazopyridine, phencyclidine, phendimetrazine, phenelzine, pheniramine, phenobarbital, phenothiazine, phensuximide, phentermine, phenylbutazone, phenylephrine, phenylpropanolamine, piperocaine, prazepam, prednisolone, primidone, probenecid, progesterone, propiomazine, propranolol, propylparaben, pseudoephedrine, puromycin, pyrilamine, pyrithyldione, quazepam, quinaldic acid, quinidine, quinine, ranitidine, recinnamine, reserpine, resorcinol, saccharin, albuterol, salicylamide, salicylic acid, scopolamine, scopoletin, secobarbital, strychnine, sulfacetamide, sufadiazine, sulfadimethoxine, sulfaethidole, sulfamerazine, sulfamethazine, sulfamethoxazole, sulfanilamide, sulfapyridine, sulfasoxazole, sulindac, tamoxifen, temazepam, testosterone, tetracaine, tetracycline, tetramisole, thebaine, theobromine, theophylline, thiabendazole, thiamine, thiamylal, thiobarbituric acid, thioridazine, thiosalicylic acid, thiothixene, thymol, tolazamide, tolazoline, tobutamide, tolmetin, tranlycypromine, triamcinolone, tribenzylamine, trichloromethiazide, trifluoperazine, trihexyphenidyl, trimethoprim, tripeleennamine, triprolidine, tropacocaine, tyramine, verapamil, vincamine, warfarin, yohimbine, zoxazolamine

REFERENCE

- Hill,D.W.; Kind,A.J. Reversed-phase solvent gradient HPLC retention indexes of drugs, *J.Anal.Toxicol.*, **1994**, *18*, 233–242.